

Currently Pending Claims

24. (Amended) A method of inducing gene expression in a mammalian cell, said method comprising:

(a) transducing the mammalian cell with (i) a first recombinant adeno-associated virus (AAV) virion comprising an AAV vector that comprises a transcriptional promoter region operably linked to a polynucleotide of interest, wherein the transcriptional promoter region comprises at least one ecdysone-responsive element (EcRE), and a promoter capable of directing the *in vivo* transcription of said polynucleotide of interest in a mammalian cell, located downstream of the at least one EcRE; and (ii) a second recombinant AAV virion comprising an AAV vector that comprises a coding sequence encoding an ecdysone receptor (EcR) and further comprises a coding sequence encoding a retinoid-X-receptor (RXR), wherein said EcR and RXR coding sequences are operably linked to control elements capable of directing the *in vivo* transcription thereof in the mammalian cell; and

(b) providing ecdysone, or an analog thereof capable of binding the EcR, to said mammalian cell, in an amount sufficient to induce expression of the polynucleotide of interest.

26. A method of inducing gene expression in a mammalian cell, said method comprising:

(a) transducing the mammalian cell with (i) a first recombinant adeno-associated virus (AAV) virion comprising an AAV vector that comprises a transcriptional promoter region operably linked to a polynucleotide of interest, wherein the transcriptional promoter region comprises five ecdysone-responsive elements positioned upstream of a heat shock protein (Hsp) promoter sequence, wherein the transcriptional promoter region is capable of directing the *in vivo* transcription of said polynucleotide of interest in a mammalian cell; (ii) a second recombinant AAV virion comprising an AAV vector that comprises a coding

sequence encoding an ecdysone receptor (EcR) operably linked to control elements capable of directing the *in vivo* transcription of said EcR coding sequence in a mammalian cell; and (iii) a third recombinant AAV virion comprising an AAV vector that comprises a coding sequence encoding a retinoid-X-receptor (RXR) operably linked to control elements capable of directing the *in vivo* transcription of said RXR coding sequence in the mammalian cell; and

(b) providing ponasterone A to said mammalian cell, in an amount sufficient to induce expression of the polynucleotide of interest.

27. The method of claim 26, wherein the transcriptional promoter region of the AAV vector of the first recombinant AAV virion further comprises at least one enhancer sequence.

28. The method of claim 27, wherein the enhancer sequence is an SP1 enhancer sequence.

29. The method of claim 27, wherein the transcriptional promoter region comprises three SP1 enhancer sequences.

30. (New) The method of claim 24, wherein the transcriptional promoter region of the AAV vector of the first recombinant AAV virion further comprises at least one enhancer sequence.

31. (New) The method of claim 30, wherein the enhancer sequence is an SP1 enhancer sequence.

32. (New) A method of inducing gene expression in a mammalian cell, said method comprising:

(a) transducing the mammalian cell with (i) a first recombinant adeno-associated virus (AAV) virion comprising an AAV vector that comprises a

transcriptional promoter region operably linked to a polynucleotide of interest, wherein the transcriptional promoter region comprises at least one ecdysone-responsive element (EcRE), and a promoter capable of directing the *in vivo* transcription of said polynucleotide of interest in a mammalian cell, located downstream of the at least one EcRE; (ii) a second recombinant AAV virion comprising an AAV vector that comprises a coding sequence encoding an ecdysone receptor (EcR) operably linked to control elements capable of directing the *in vivo* transcription thereof in the mammalian cell; and (iii) a third recombinant AAV virion comprising an AAV vector that comprises a coding sequence encoding a retinoid-X-receptor (RXR) operably linked to control elements capable of directing the *in vivo* transcription thereof in the mammalian cell; and

(b) providing ecdysone, or an analog thereof capable of binding the EcR, to said mammalian cell, in an amount sufficient to induce expression of the polynucleotide of interest.

33. (New) The method of claim 32, wherein the transcriptional promoter region of the AAV vector of the first recombinant AAV virion further comprises at least one enhancer sequence.

34. (New) The method of claim 33, wherein the enhancer sequence is an SP1 enhancer sequence.

35. (New) A method of inducing gene expression in a mammalian cell, said method comprising:

(a) transducing a mammalian cell comprising a retinoid-X-receptor (RXR) with (i) a first recombinant adeno-associated virus (AAV) virion comprising an AAV vector that comprises a transcriptional promoter region operably linked to a polynucleotide of interest, wherein the transcriptional promoter region comprises at least one ecdysone-responsive element (EcRE), and a promoter capable of

directing the *in vivo* transcription of said polynucleotide of interest in a mammalian cell, located downstream of the at least one EcRE and (ii) a second recombinant AAV virion comprising an AAV vector that comprises a coding sequence encoding an ecdysone receptor (EcR) operably linked to control elements capable of directing the *in vivo* transcription thereof in the mammalian cell; and

(b) providing ecdysone, or an analog thereof capable of binding the EcR, to said mammalian cell, in an amount sufficient to induce expression of the polynucleotide of interest.

36. (New) The method of claim 35, wherein the transcriptional promoter region of the AAV vector of the first recombinant AAV virion further comprises at least one enhancer sequence.

37. (New) The method of claim 36, wherein the enhancer sequence is an SP1 enhancer sequence.